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Response of White Leghorn Chickens of Various *B* Haplotypes to Infection at Hatch with Subgroup J Avian Leukosis Virus

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SUMMARY. White leghorn chickens from seven 15.*B* congenic lines (genetically similar except for genes linked to the major histocompatibility complex [MHC] *B* haplotype) and two Line 0.*B* semicongenic lines were infected at hatch with strain ADOL Hc-1 of subgroup J avian leukosis virus (ALV-J). At 5, 8, 16, and 36 wk of age, chickens were tested for viremia, serum-neutralizing antibody, and cloacal shedding. Chickens were also monitored for development of neoplasia. In the 15.*B* congenic lines (*B*2, B*5, B*12, B*13, B*15, B*19*, and *B*21*) there were no significant differences in the incidence of viremia between *B* haplotypes. In fact, infection at hatch in all of the 15.*B* congenic lines induced tolerance to ALV-J because 100% of these chickens were viremic and transient circulating serum-neutralizing antibody was detected in only a few chickens throughout the 36 wk experiment. However, at 16 wk of age more *B*15* chickens had antibody and fewer *B*15* chickens shed virus than did the 16-wk-old *B*2, B*5*, or *B*13* chickens. Moreover, compared with *B*15* chickens, a higher percentage of *B*13* chickens consistently shed virus from 8 wk postinfection to termination at 36 wk postinfection. The *B* haplotype had a transient effect on viral clearance in Line 0.*B* semicongenics, as more *B*13* than *B*21* chickens remained viremic through 5 wk of age. Very few (0%–18%) of the Line 0.*B* semicongenic chickens shed virus. By 36 wk of age, all Line 0 *B*13* and *B*21* chickens produced serum-neutralizing antibodies and cleared the virus. These results show that following ALV-J infection at hatch the immune response is influenced transiently by the *B* haplotype and strongly by the line of chicken. Although this study was not designed to study the effect of endogenous virus on ALV-J infection, the data suggest that endogenous virus expression reduced immunity to ALV-J in Line $15I_5$, compared with Line 0, a line known to lack endogenous virus genes.

RESUMEN. Respuesta de aves leghorn blancas de varios haplotipos *B* a la infección al nacimiento con virus de Leucosis aviar subgrupo J.

Utilizando la cepa ADOL Hc-1 del virus de leucosis aviar subgrupo J se infectaron al nacimiento aves leghorn blancas provenientes de siete líneas genéticas congénicas 15.*B* (genéticamente similares excepto por los genes ligados al complejo mayor de histocompatibilidad del haplotipo B) y dos líneas semicongénicas 0.*B*. Las aves fueron evaluadas para determinar viremia, anticuerpos neutralizantes en suero y dispersión viral por cloaca a las 5, 8, 16, y 36 semanas de edad. A su vez se evaluó el desarrollo de neoplasias en las aves. En las líneas congénicas 15.*B* (*B*2, B*5, B*12, B*13, B*15, B*19*, y *B*21*) no se observaron diferencias significativas en la incidencia de viremia entre haplotipos *B*. De hecho, la infección al nacimiento indujo tolerancia al virus de leucosis aviar subgrupo J en todas las líneas congénicas 15.*B,* debido a que en estas aves hubo un 100% de viremia, detectándose de manera transitoria anticuerpos séricos neutralizantes sólo en unas pocas aves durante las 36 semanas del experimento. Sin embargo, a las 16 semanas de edad más aves *B*15* presentaron anticuerpos y menos aves del mismo grupo dispersaron virus que las aves *B*2, B*5,* o *B*13.* Por otra parte, comparados con las aves *B*15*, un mayor porcentaje de aves *B*13* dispersaron virus consistentemente desde la octava semana post infección hasta la semana 36 post infección. El haplotipo *B* generó un efecto transitorio en la eliminación viral en la línea semioncogénica 0.*B,* puesto que más aves *B*13* que las *B*15* persistieron virémicas hasta la quinta semana de edad. Muy pocas aves de la línea 0.*B* (0%–18%) dispersaron virus. Para la semana 36 todas las aves de las líneas 0.*B*13* y *B*21* produjeron anticuerpos séricos neutralizantes y eliminaron completamente el virus. Estos resultados demuestran que posterior a una infección al nacimiento con virus de leucosis aviar subgrupo J, la respuesta inmune se ve influenciada transitoriamente por el haplotipo *B* y fuertemente por la línea genética del ave. Sin embargo, este estudio no fue diseñado para evaluar el efecto de virus endógenos sobre la infección con virus de leucosis aviar subgrupo J. Los datos sugieren que la expresión de virus endógenos redujo la inmunidad contra el virus de leucosis aviar subgrupo J en la línea $15I_5$ al comparase con la línea 0, la cual es conocida por no poseer genes de virus endógenos.

Key words: avian leukosis virus, *B* haplotype, white leghorn, chickens, genetic effects

Abbreviations: ALVE = endogenous avian leukosis virus; ALV-A = subgroup A avian leukosis virus; ALV-J = subgroup J avian leukosis virus; *EV* = endogenous viral gene; MD = Marek's disease; MHC = major histocompatibility complex; TCID = tissue culture infectious dose

Avian leukosis virus subgroup J (ALV-J) causes myeloid leukosis, predominantly in meat-type chickens (27,43,45) and infrequently in egg-type chickens (44). ALV-J has caused severe economic losses in broiler breeders. Vertical transmission from broiler breeders to progeny is more frequent with ALV-J than with other ALV subgroups (57). Broiler breeder companies have instituted expensive programs to reduce or eliminate ALV-J infection in elite breeding stock by identification and removal of dams that are likely to vertically shed virus to progeny chicks (41). However, identification of transmitter dams in broilers is difficult and even rigorous testing may not identify all transmitter dams (57). In addition, genetic factors involved in resistance to ALV-J infection have not been identified. This makes eradication and control of ALV-J very difficult.

The genes closely linked to the polymorphic *EaB* blood group locus (18) compose the major histocompatibility complex (MHC) (49).

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The chicken MHC genes in the *B* haplotype include a complex of two Class I loci (*BF1* and *BF2*), two Class II loci (*BL1* and *BL2*), and Class IV (*BG*) genes (40). The Class I (*BF*) and Class II (*BL*) genes are similar to the MHC genes of mammalian species. The *BG* genes are unique to avian species and are expressed on red blood cells. The antigenic BG molecules are the major epitopes identified by hemagglutination (30). Genes linked to the MHC *B* haplotype have been associated with disease resistance and vaccinal immunity in chickens (2,7,8,9,35).

White Leghorn lines have been shown to differ in immunity to ALV-J. For example, after infection at hatch with ALV-J, 93% of Line 0 (*B*21*) chickens had detectable serum-neutralizing antibody by 30 wk, in contrast to 43% of Line $15I_5$ (*B*15*) chickens (56). However, the possible influence of the *B* haplotype on this line difference has not been tested in *B* congenic lines within either Line 0 or Line $15I_5$. In pilot studies the *B* haplotype influenced the time of ALV-J clearance following multiple infections of 1-to-2-yr-old 15.*B* congenic hens (3). No studies have been conducted in *B* congenic lines within either Line $15I_5$ or Line 0 infected with ALV-J at hatch. The depression of immune response to ALV-J due to the expression of endogenous viral genes (*EV*) has been described previously in Line 0.*EV* congenic lines (6,56). The current experiments were done with chickens that express endogenous virus and have a slow immune response to ALV-J ($15I_5$ and six *B* congenic lines) and with chickens that lack endogenous virus and have a rapid immune response to ALV-J (Line 0 and one *B* semi-congenic line).

MATERIALS AND METHODS

Chickens. *B* congenic chickens from two lines developed in this laboratory were used: 15.*B* congenic lines and 0.*B* semicongenic lines. The 15.*B* congenic lines were developed in inbred white leghorn Line $15I_5$ in order to clarify the role of individual *B* haplotype genes in disease resistance (2). The development of the 15.*B* congenic lines involved 10–11 generations of backcross matings to inbred Line $15I_5$ (2,6,50). Each of the 15.*B* congenic lines is 99.9% identical to the inbred parental Line $15I_5$, but is homozygous for different genes in the *B* haplotype (6). Two of the 15.*B* congenic lines are homozygous for the *B*2* haplotype introduced from Marek's disease (MD)-resistant (Line 6_3) or MD-susceptible (Line 7_2) chickens. The remaining 15.*B* congenic lines are homozygous for *B*5, B*12, B*13, B*15* ($15I_5$), *B*19*, or *B*21.*

Two additional *B* semicongenic lines were developed in Line 0 (6). Line 0 was developed by producing F_1 chickens from Line $7_2 \times$ SPAFAS Line 11 mating and then backcrossing to Line 11 (1). Line 0 was then selected for absence of endogenous avian leukosis virus genes (*ALVE*) (1), and subsequently for resistance to subgroup E avian leukosis virus (ALVE) (48). Noninbred Line 0 initially segregated for several *B* genes; however, this was fixed for the *B*21* haplotype (Crittenden and Bacon, unpubl. data). Line 0.P-*13* was developed by introducing the *B*13* haplotype from the 15.*B* congenic line 15.P-*13* into Line 0 (H. Hunt, J. Dodgson, L. Bacon, unpubl. data). Line 0.P-*13* is reproduced each year by mating *B*13/*21* males to Line 0 hens and selecting for *B*13/*21* breeders. The *B*13/*21* breeders are mated to produce *B* haplotype homozygous and heterozygous experimental chicks. Lines 0-*21* and 0.P-*13* are important for analysis of the role of the *B* haplotype in resistance to an infection in the absence of *ALVE* expression. In contrast, the effect of *B* haplotype in the 15.*B* congenic lines occurs in the presence of expression from *ALVE* genes *1, 6, 10*, and *11*(6).

Twenty-five chickens were obtained from each of seven 15.*B* congenic lines: $15I_5$ (*B*15*), 15.7-*2*, 15.15I-*5*, 15.C-*12*, 15.P-*13*, 15.P-*19*, and 15.N-*21.* The eighth 15.*B* congenic line, 15.6-*2*, was not included since this line is 99.9% identical to line 15.7-*2.* Sixty-four chickens were obtained from 0.P-*13* matings of *B*13/*21* males and females. These chickens randomly possessed the *B*13/*13, B*13/*21*, or *B*21/*21* genotypes. The breeder chickens were monitored for freedom from pathogens as described previously (53).

Virus. Passage seven of ALV-J strain ADOL-Hc1 (27) from this laboratory was used to infect chicks at 1 day of age.

Virological and antibody assays. Plasma and cloacal swabs from inoculated chickens were tested for ALV as described (28). Briefly, samples were inoculated on Line 0 (resistant to ALVE) chicken embryo fibroblasts (22). Seven to 9 days later, cell lysates were tested for the presence of ALV group-specific antigen by an enzyme-linked immunosorbent assay (52). Serum virus neutralizing antibody response was determined by neutralization tests (28).

Definition of *B* haplotypes by hemagglutination. The *B* haplotypes of the 0.*B* semicongenic chickens were defined by hemagglutination using *B*13* and *B*21* specific alloantisera (30).

Pathology. All chickens were necropsied. Tissues with evidence of tumors were fixed in 10% buffered formalin, stained with hematoxylin and eosin, and examined for microscopic lesions. ALV-J-induced neoplasia was diagnosed based on characteristic gross and microscopic lesions (25).

Statistical analysis. The test statistic was chi-square analysis performed using SAS statistical software for Windows, version 8.2 (SAS Institute Inc., Cary, NC).

Experimental design. All chickens were hatched at one time. The chickens were inoculated intra-abdominally with 10^4 tissue culture infectious dose (TCID)/ml ADOL-Hc1 at hatch based upon previous experiments (56). Chickens of all genotypes were intermingled in colony cages within the same room. At 23 wk of age half the male chickens of each 15.*B* congenic line, and the line 0 *B*13/*21* genotype, were bled and terminated to make more cages available. The remaining chickens were placed into individual male and female adult cages within the same room. Chickens were tested at 5, 8, 16, and 36 wk of age for ALV-J virus in serum and cloacal swabs and for antibody.

RESULTS

Blood typing. Sixty-four 0.*B* semicongenic chickens were blood-typed at 2 wk of age. Thirteen chickens were *B*13/*13* homozygotes, 31 were *B*13/*21* heterozygotes, and 20 were *B*21/*21* homozygotes. This frequency of homozygotes to heterozygotes is within the expected genotype frequency of 1:2:1. At 23 wk of age seven excess *B*13/*21* males were terminated. A total of seven *B*13/*13*, 15 *B*13/*21*, and 15 *B*21/*21* chickens remained until termination at 36 wk of age.

Effect of MHC *B* haplotype on ALV-J-induced viremia and serum-neutralizing antibody. *15.B congenic lines.* Infection of the 15.*B* congenic lines with ALV-J at hatch resulted in viremic tolerant (V^+A^-) chickens at 36 wk postinfection. In all tests 91%–100% of the 15.*B* congenic chickens of all lines were viremic (Table 1). In addition, all excess males terminated at 23 wk of age were viremic tolerant regardless of *B* haplotype (data not shown). Serum-neutralizing antibodies against ALV-J were detected in plasma of a small percentage (9%–20%) of $15I_5$ (*B*15*), 15.P-*19* (*B*19*), 15.N-*21* (*B*21*), and 15.C-*12* (*B*12*) chickens at 16 wk postinfection (Table 2). At this age, 20% of the Line $15I_5$ (*B*15*) chickens produced antibody against ALV-J whereas 0% of chickens from each of the lines 15.7-*2* (*B*2*), 15.15I-*5* (*B*5*), and 15.P-*13* (*B*13*) produced serum-neutralizing antibodies ($P < 0.05$). The level of circulating serum-neutralizing antibody was transient and insufficient to clear the ALV-J.

0.B semicongenic lines. Viremia was detected in a higher percentage of *B*13/*13* and *B*13/*21* than *B*21/*21* chickens at 5 wk postinfection ($P < 0.05$) (Table 1). At 5 wk, 64% of the *B*13/*13* chickens remained viremic as compared to 38% *B*13/*21* and 11% of *B*21/*21* chickens ($P < 0.05$). However, by 8 wk the *B* haplotype no longer influenced the viremia status and at 16 wk only one chicken (*B*13/*21*) remained viremic. There were no significant differences in development of serum-neutralizing antibodies between the different 0.*B* semicongenic chickens following ALV-J infection at hatch (Table 2). All birds within the 0.*B* semicongenic lines had produced antibody and were either nonviremic immune (V^-A^+) or lacking both virus

Table 1. Virus isolation from plasma samples from 15.*B* congenic and 0.*B* semicongenic chickens following infection at hatch.[AB]

Line	*B* haplotype	5 wks PI[C]	8 wks PI	16 wks PI	36 wks PI
15.*B* congenic	*B*2*	20/21 (95%)[a]	21/21 (100%)[a]	21/21 (100%)[a]	17/17 (100%)[a]
	*B*5*	22/22 (100%)[a]	22/22 (100%)[a]	22/22 (100%)[a]	18/18 (100%)[a]
	*B*12*	25/25 (100%)[a]	24/25 (96%)[a]	25/25 (100%)[a]	20/20 (100%)[a]
	*B*13*	25/25 (100%)[a]	25/25 (100%)[a]	25/25 (100%)[a]	20/20 (100%)[a]
	*B*15*	24/25 (96%)[a]	25/25 (100%)[a]	23/25 (92%)[a]	22/22 (100%)[a]
	*B*19*	24/24 (100%)[a]	22/24 (92%)[a]	23/23 (100%)[a]	19/19 (100%)[a]
	*B*21*	24/24 (100%)[a]	23/24 (96%)[a]	21/23 (91%)[a]	17/17 (100%)[a]
0.*B* semicongenic	*B*13/*13*	7/11 (64%)[c]	1/8 (12%)[b]	0/7 (0%)[b]	0/7 (0%)[b]
	*B*13/*21*	10/26[D] (38%)[c]	1/27 (4%)[b]	1/25 (4%)[b]	0/15 (0%)[b]
	*B*21/*21*	2/18 (11%)[b]	0/17 (0%)[b]	0/18 (0%)[b]	0/15 (0%)[b]

[A]All chickens were infected with strain ADOL Hc1 of ALV-J at 1 day of age. The presence of virus in plasma samples was determined as previously described (27).
[B]Within each weekly column the percentages within each *B* congenic line series with different lowercase letters differ significantly based upon chi-square analyses ($P < 0.05$).
[C]PI = postinfection.
[D]At 5 weeks of age, one chicken yielded insufficient plasma sample and was not tested.

and antibody (V^-A^-) at 36 wk postinfection. Thus, in Line 0.*B* semicongenic chickens lacking endogenous virus, the *B* haplotype influenced the duration of ALV-J infection. This influence was transient and did not persist at or beyond 8 wk. The clearance of virus in 0.*B* semicongenic chickens was unlike that of Line $15I_5$ congenic chickens, where all the birds remained viremic through 36 wk.

Effect of MHC *B* haplotype on ALV-J cloacal shedding. In the 15.*B* congenic chickens the *B* haplotype influenced cloacal shedding. The percentage of $15I_5$ (*B*15*) chickens shedding virus at 8, 16, and 36 wk of age was consistently lower than some of the other *B* haplotypes (Table 3). Alternatively, the percentage of virus-shedders in line 15.P-*13* (*B*13*) was consistently high at all ages.

In the 0.*B* semicongenic lines, the *B* haplotype did not influence cloacal shedding at any age (Table 3).

Tumor susceptibility. Very few chickens developed neoplasia following infection with ALV-J at hatch. The *B* haplotype did not influence tumor susceptibility in the 15.*B* congenic or 0.*B* semicongenic chickens (Table 4).

DISCUSSION

In the 15.*B* congenic chickens infected at hatch, the incidence of ALV-J-induced viremia was comparable in all lines (Table 1). All the chickens were persistently viremic and at 36 wk of age, only one (a *B*15* chicken) of 133 chickens had antibody; the rest were viremic tolerant. However, the *B* haplotype did have a significant influence on cloacal shedding (Table 3), as well as on transient production of serum-neutralizing antibody (Table 2). The percentage of the *B*13* chickens shedding virus was among the highest of all *B* genotypes from 5 to 36 wk. Alternatively, from 8 to 36 wk postinfection a lower percentage of *B*15* chickens were shedding virus compared to some of the other 15.*B* congenic lines. The production of serum-neutralizing antibodies was seen in a small percentage of the *B*15, B*19, B*21,* and *B*12* chickens at 16 wk after infection, although the level of antibody was insufficient to clear the ALV-J virus and these birds remained viremic tolerant. However, at 16 wk after infection at hatch, a higher percentage of *B*15* chickens than *B*2, B*5*, or *B*13* chickens produced antibody (Table 2). The antibody and shedding data suggest that *B*15* chickens are more competent to respond to ALV-J than are *B*13* 15.*B* congenic chickens.

The MHC *B* haplotype influenced ALV-J viremia following infection at hatch in Line 0.*B* semicongenic chickens, which lack endogenous ALV. Transient differences were seen in viremia between different *B* genotypes. For example, more *B*21/*21* chickens mounted a virus-eradicating immune response and cleared the ALV-J virus by 5 wk postinfection, whereas most *B*13/*13* homozygotes did not clear the virus until 8 wk postinfection (Table

Table 2. Serum-neutralizing antibody following infection of 15.*B* congenic and 0.*B* semicongenic chickens at hatch.[AB]

Line	*B* haplotype	5 wks PI[C]	8 wks PI	16 wks PI	36 wks PI
15.*B* congenic	*B*2*	3/21 (14%)[a,b]	1/21 (5%)[a]	0/21 (0%)[a]	0/17 (0%)[a]
	*B*5*	0/22 (0%)[a]	0/22 (0%)[a]	0/22 (0%)[a]	0/18 (0%)[a]
	*B*12*	0/25 (0%)[a]	0/25 (0%)[a]	1/25 (4%)[a,b]	0/20 (0%)[a]
	*B*13*	0/25 (0%)[a]	0/25 (0%)[a]	0/25 (0%)[a]	0/20 (0%)[a]
	*B*15*	8/25 (32%)[b]	0/25 (0%)[a]	5/25 (20%)[b]	1/22 (5%)[a]
	*B*19*	0/24 (0%)[a]	0/24 (0%)[a]	3/23 (13%)[a,b]	0/19 (0%)[a]
	*B*21*	0/24 (0%)[a]	1/24 (4%)[a]	2/23 (9%)[a,b]	0/17 (0%)[a]
0.*B* semicongenic	*B*13/*13*	4/11 (36%)[c]	3/8 (38%)[c]	3/7 (43%)[c]	5/7(71%)[d]
	*B*13/*21*	6/26 (23%)[c]	6/27 (22%)[c]	13/25 (52%)[c]	4/15 (27%)[c]
	*B*21/*21*	4/18 (22%)[c]	4/17 (24%)[c]	11/18 (61%)[c]	8/15 (53%)[c,d]

[A]All chickens were infected with strain ADOL Hc1 of ALV-J at 1 day of age. The presence of serum-neutralizing antibody in plasma samples was determined as previously described (27).
[B]Within each weekly column the percentages within each *B* congenic line series with different lowercase letters differ significantly based upon chi-square analyses ($P < 0.05$).
[C]PI = postinfection.

Table 3. Cloacal shedding in 15.*B* congenic and 0.*B* semicongenic chickens following infection at hatch.[AB]

Line	*B* haplotype	5 wks PI[C]	8 wks PI	16 wks PI	36 wks PI
15.*B* congenic	*B*2*	16/21 (76%)[a]	10/21 (48%)[a]	10/21 (48%)[a]	16/17 (94%)[a,b]
	*B*5*	21/22 (95%)[a,b]	11/22 (50%)[a]	12/22 (55%)[a,b]	18/18 (100%)[b]
	*B*12*	25/25 (100%)[b]	15/25 (60%)[a]	12/25 (48%)[a]	20/20 (100%)[b]
	*B*13*	25/25 (100%)[b]	24/25 (96%)[b]	19/25 (76%)[b]	19/20 (90%)[a,b]
	*B*15*	24/25 (96%)[b]	12/25 (48%)[a]	8/25 (32%)[a]	17/22 (77%)[a]
	*B*19*	21/24 (88%)[a,b]	14/24 (58%)[a]	12/23 (52%)[a,b]	17/19 (89%)[a,b]
	*B*21*	22/24 (92%)[a,b]	11/24 (46%)[a]	15/23 (65%)[a,b]	15/17 (88%)[a,b]
0.*B* semicongenic	*B*13/*13*	2/11 (18%)[c]	1/8 (12%)[c]	0/7 (0%)[c]	0/7 (0%)[c]
	*B*13/*21*	2/26 (8%)[c]	1/27 (4%)[c]	0/25 (0%)[c]	1/15 (7%)[c]
	*B*21/*21*	2/18 (11%)[c]	0/17 (0%)[c]	1/18 (5%)[c]	1/15 (7%)[c]

[A]All chickens were infected with strain ADOL Hc1 of ALV-J at 1 day of age. The presence of virus in cloacal swabs was determined as previously described (27).

[B]Within each weekly column the percentages within each *B* congenic line series with different lowercase letters differ significantly based upon chi-square analyses ($P < 0.05$).

[C]PI = postinfection.

1). We conclude that, compared to *B*21* chickens, *B*13* chickens appear less immunoresponsive to ALV-J in both Line 0 and Line $15I_5$. This decrease in immune response in *B*13* chickens may be attributed to the cytotoxic T lymphocyte (CTL) response. Adult chickens with the *B*13* haplotype show little to no CTL activity following infection with subgroup A avian leukosis virus (ALV-A), whereas chickens with the *B*21* haplotype show high CTL activity (55). Induction of a CTL response is dependent on the presentation of viral antigens complexed with MHC glycoproteins on the cell surface.

The lower immune responsiveness of *B*13* chickens to ALV-J in contrast to *B*21* chickens in the Line 0 or *B*15* chickens in the Line $15I_5$ background lines is in agreement with earlier studies on ALV-A infection in the $15I_5$ *B* congenic lines. Following infection at 1–2 wk of age with ALV-A, over 76% of the chickens in all lines developed lymphoid leukosis. However, serum-neutralizing antibody was only detected in 26% of *B*13* chickens in contrast to over 77% of *B*2, B*5, B*12, B*15*, and *B*19* chickens (*B*21* was not available) (5,10).

Table 4. Percentage of 15.*B* congenic and 0.*B* semicongenic chickens developing tumors by 36 wks of age following infection with ALV-J at hatch.[AB]

Line	*B* haplotype	Number of birds with neoplasia/total number of birds at risk (%)
15.*B* congenic	*B*2*	2/25 (8%)
	*B*5*	3/24 (12%)
	*B*12*	0/25 (0%)
	*B*13*	2/23 (9%)
	*B*15*	0/25 (0%)
	*B*19*	2/25 (8%)
	*B*21*	2/25 (8%)
0.*B* semicongenic	*B*13/*13*	0/13 (0%)
	*B*13/*21*	3/32 (9%)
	*B*21/*21*	1/19 (5%)

[A]Number of chickens with neoplasia/total number of chickens at risk (percentage). The total includes six to eight males of each 15.*B* congenic line and the seven *B*13/*21* males terminated at 23 wks of age.

[B]Types of neoplasia included hemangioma, erythroblastosis, multihistiocytic sarcoma, rhabdosarcoma, and renal and gonadal tumors. Based upon chi-square analyses there were no significant differences in the percentage of tumors between lines within each *B* congenic line series ($P > 0.05$).

In addition, following Rous sarcoma virus inoculation of RSV(RAV-1) the 15.*B* congenic chickens with *B*13, B*5, B*15*, and *B*19* haplotypes developed tumors that grew rapidly and often metastasized, whereas tumors in chickens with *B*2, B*12* and *B*21* haplotypes regressed and metastasis was infrequent (4,6). In regard to other viral pathogens, the *B*13* haplotype confers susceptibility to Marek's disease virus induced tumors, whereas the *B*21* haplotype confers resistance (2). Recently, *B*13* and *B*21* chickens were shown to be less responsive to a lowly attenuated infectious bronchitis virus vaccine when compared to *B*15* chickens (7). We conclude that 15.*B* congenic chickens with the *B*13* haplotype are less responsive than other *B* haplotypes to an array of poultry pathogens and vaccines. Interestingly, *B*13* is common in White Leghorns (17) and broilers (37). In experiments using broiler sublines fixed for different *B* haplotypes, no differences were seen in susceptibility to infectious bursal disease between genotypes *B*13/*13, B*13/*21*, and *B*21/*21* (33). However, in response to an infectious *E. coli, B*13/*13* broilers and leghorns were more resistant to cellulitis than were *B*21/*21* broilers or leghorns (38,39). Thus, the *B*13* haplotype may confer protection against some pathogens.

The background genes of Lines 0 and $15I_5$ differ in many ways. However, one explanation for the difference in the ability to clear ALV-J virus between the 15.*B* congenics and 0.*B* semicongenics is expression of *ALVE* genes in the 15.*B* congenic lines. The 0.*B* semicongenic lines were selected for absence of all *ALVE* genes (1) and are resistant to ALVE infection due to the absence of an *ALVE* receptor (48). In contrast, the 15.*B* congenic lines are susceptible at the receptor level to endogenous virus infection and contain *ALVE1, 6, 10,* and *11* genes (6). The *ALVE1* gene is very common among White Leghorn chickens and expresses little to no detectable viral protein product (19,46,54). The *ALVE6* gene lacks the 5′ LTR and *gag* sequences but the rest of the provirus is intact, producing normal envelope glycoproteins (11,32). The genes *ALVE10* and *11* produce complete virus in the presence of *ALVE1* (20,21). It is well established that the presence of *ALVE* gene expression increases susceptibility to infection with exogenous ALV (23,24,26,51). Moreover, this laboratory has shown that the expression of the *ALVE21* gene (linked to slow feathering) in Line 0 semicongenic chickens results in increased viremia and reduced immune response to ALV-J following infection at hatch (56). This is the most compelling evidence that *ALVE* gene expression leads to immunological tolerance after ALV-J infection. Numerous *ALVE* genes have been characterized in chickens; some of these genes are unique to meat-type chickens (13,14,15,31). We conclude that tolerance or immunity is greatly influenced by *EV* genes in Line $15I_5$ as opposed to Line 0. The timing of testing determines the degree of tolerance or immunity detected in theses lines (56).

There was no significant difference in development of neoplasia in white leghorn chickens with different *B* genotypes following ALV-J infection at hatch. None of the 15.*B* congenic and 0.*B* semicongenic chickens developed myeloid leukosis. However, a small percentage developed hemangiomas, erythroblastosis, multihistiocytic sarcomas, rhabdosarcomas, and renal tumors. Similarly, leukosis/sarcoma tumors, but not myeloid leukosis, were detected in $15I_5$ chickens infected with ALV-J strain HPRS-103 at hatch (44,45). Myeloid leukosis and/or renal tumors have been observed in Line 0 but at lower frequencies than observed in meat-type lines (42). The type of ALV-induced neoplasia is influenced by the strain of virus, exposure dose, route, and host genotype, sex, and age at exposure (25).

The age at infection is known to affect development of serum-neutralizing antibodies and retention of ALV-J. In this study with $15I_5$ chickens infected at hatch, viremia was persistent and only transient antibody developed. In a pilot study with aged hens given multiple injections of cells infected with ALV-J, the 15.*B* congenic chickens developed high titers of serum-neutralizing antibodies, with the actual levels varying, depending on the *B* genotype (3). In that study, the majority of *B*15* and *B*21* hens cleared virus by 3 wk after the first immunization, whereas *B*2* and *B*5* hens did not clear ALV-J until 6 to 12 wk postimmunization. Furthermore, some *B*13* hens did not produce sufficient serum-neutralizing antibodies to clear the virus until after 12 wk postimmunization. In this experiment a smaller percentage of $15I_5$ chickens had detectable serum-neutralizing antibody at 36 wk of age than was observed at 30 wk of age by Williams *et al.* (56). ALV-J infection occurred at hatch in both experiments. This reduced level of antibody response may be attributed to a greater number of viremic tolerant chickens that persistently shed virus, reinfecting chickens in the same cages in this experiment as compared to the earlier study. Payne *et al.* (42) previously observed horizontal transmission between viremic tolerant and noninfected broiler chickens, but not between white leghorns.

The effects of genes linked to the MHC *B* haplotype on ALV-J infection and tumor development may be more significant clinically in meat-type chickens, in which ALV-J causes predominantly myeloid leukosis (45). However, the MHC *B* haplotypes in meat-type strains are only recently becoming defined. Interestingly, some broiler strains possess *BF* and *BL* genes identical to genes in white leghorns of the *B*2, B*12, B*13, B*15*, and *B*21* haplotypes (36,37). Unfortunately, there are no *B* congenic lines developed for meat-type chickens and we have not been able to acquire broilers of defined *B* genotypes for infection with ALV-J (A. Pandiri, pers. comm.). Commercial broiler breeder lines are derived from separate male and female lines, resulting in increased variability in disease resistance between breed crosses (34). Based on our results in white leghorn chickens, we suspect that some of the variability in immune response to ALV-J in broilers may result from differences in expression of *ALVE* genes, as well as differences in genes linked to the *B* haplotype. In fact, meat-type chickens frequently contain more *EV* genes than do egg-type chickens (16,47). Further improvements in the identification of *ALVE* loci (12) and *ALVE* receptor genes (58), as well as *B* haplotypes (29,36,37,40), may soon permit evaluation of these genes for resistance to disease in commercial egg-laying and broiler chicken strains.

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